

AMENDMENTS TO THE CLAIMS

1-22. (Cancelled)

23. (New) A method for detecting a single nucleotide polymorphism in a target comprising, under isothermal conditions at about 37 degrees Celsius:

- a) hybridizing a detector primer and a second primer to the target such that extension of the second primer by polymerase displaces the detector primer from the target sequence, wherein the detector primer comprises a diagnostic nucleotide for the single nucleotide polymorphism which is one to four nucleotides from the 3' terminal nucleotide of the detection primer;
- b) extending the detector primer and the second primer with polymerase to produce a displaced detector primer extension product;
- c) determining an efficiency of detector primer extension; and
- d) detecting the presence or absence of the single nucleotide polymorphism based on the efficiency of detector primer extension.

24. (New) The method of Claim 23 wherein the single nucleotide polymorphism is identified using the detector primer.

25. (New) The method of Claim 24 wherein the single nucleotide polymorphism is identified using multiple detector primers, each comprising a different diagnostic nucleotide.

26. (New) The method of Claim 25 wherein two detector primers are used to identify which of two possible alleles is present in the target sequence.

27. (New) The method of Claim 25 wherein four detector primers are used to identify the nucleotide present in the target sequence at the position of the single nucleotide polymorphism.

28. (New) The method of Claim 25 wherein each of the multiple detector primers has a different 5' tail sequence.

29. (New) The method of Claim 23 wherein the detector primer further comprises a nucleotide which forms a nondiagnostic mismatch with the target sequence.
30. (New) The method of Claim 29 wherein the nondiagnostic nucleotide is positioned within fifteen nucleotides of the diagnostic nucleotide in the detector primer.
31. (New) The method of Claim 30 wherein the nondiagnostic nucleotide is positioned 1-5 nucleotides from the diagnostic nucleotide in the detector primer.
32. (New) The method of Claim 31 wherein the nondiagnostic nucleotide is adjacent to the diagnostic nucleotide in the detector primer.
33. (New) The method of Claim 29 wherein the detector primer is about 15-36 nucleotides long.
34. (New) The method of Claim 33 wherein the detector primer is about 18-24 nucleotides long.
35. (New) The method of Claim 23 wherein the second primer is an amplification primer.
36. (New) The method of Claim 35 wherein the amplification reaction is selected from the group consisting of SDA, 3SR, NASBA, and TMA.
37. (New) The method of Claim 23 wherein the detector primer is about 12-50 nucleotides long.
38. (New) The method of Claim 37 wherein the detector primer is about 12-24 nucleotides long.
39. (New) The method of Claim 38 wherein the detector primer is about 12-19 nucleotides long.

40. (New) The method of Claim 23 wherein the presence or absence of the single nucleotide polymorphism is detected by means of a label associated with the detector primer.

41. (New) The method of Claim 40 wherein the label becomes detectable upon extension of the detector primer or produces a change in signal upon extension of the detector primer.

42. (New) The method of Claim 41 wherein the label is a fluorescent donor/quencher dye pair and an increase in donor dye fluorescence is detected as an indication of the presence of the single nucleotide polymorphism.

43. (New) The method of Claim 41 wherein a change in fluorescence polarization is detected as an indication of the presence of the single nucleotide polymorphism.

44. (New) The method of Claim 23 wherein a single nucleotide polymorphism in an HFE gene is detected.

45. (New) The method of Claim 44 wherein the single nucleotide polymorphism is detected in exon 4 or exon 2 of the HFE gene.

46. (New) The method of Claim 23 wherein the efficiency of detector primer extension is determined quantitatively.

47. (New) A method for detecting a single nucleotide polymorphism in a target comprising, in an isothermal nucleic acid amplification reaction at about 37 degrees Celsius:

- a) hybridizing a detector primer to the target, wherein the detector primer comprises a diagnostic nucleotide for the single nucleotide polymorphism one to four nucleotides from a 3' terminal nucleotide of the detector primer which is complementary to the target sequence;
- b) amplifying the target by hybridization and extension of the detector primer;
- c) determining an efficiency of detector primer extension, and;

d) detecting the presence or absence of the single nucleotide polymorphism based on the efficiency of detector primer extension.

48. (New) The method of Claim 47 wherein the single nucleotide polymorphism is identified using the detector primer.

49. (New) The method of Claim 48 wherein the single nucleotide polymorphism is identified using two or more detector primers, each comprising a different diagnostic nucleotide.

50. (New) The method of Claim 49 wherein two detector primers are used to identify which of two possible alleles is present in the target sequence.

51. (New) The method of Claim 49 wherein four detector primers are used to identify the nucleotide present in the target sequence at the position of the single nucleotide polymorphism.

52. (New) The method of Claim 49 wherein each of the multiple detector primers has a different 5' tail sequence.

53. (New) The method of Claim 47 wherein the detector primer further comprises a nucleotide which forms a nondiagnostic mismatch with the target sequence.

54. (New) The method of Claim 53 wherein the nondiagnostic nucleotide is positioned within fifteen nucleotides of the diagnostic nucleotide in the detector primer.

55. (New) The method of Claim 54 wherein the nondiagnostic nucleotide is positioned 1-5 nucleotides from the diagnostic nucleotide in the detector primer.

56. (New) The method of Claim 55 wherein the nondiagnostic nucleotide is adjacent to the diagnostic nucleotide in the detector primer.

57. (New) The method of Claim 53 wherein the detector primer is about 15-36 nucleotides long.
58. (New) The method of Claim 57 wherein the detector primer is about 18-24 nucleotides long.
59. (New) The method of Claim 47 wherein the isothermal amplification reaction is selected from the group consisting of SDA, 3SR, NASBA and TMA.
60. (New) The method of Claim 47 wherein the detector primer is about 12-50 nucleotides long.
61. (New) The method of Claim 60 wherein the detector primer is about 12-24 nucleotides long.
62. (New) The method of Claim 61 wherein the detector primer is about 12-19 nucleotides long.
63. (New) The method of Claim 47 wherein the presence or absence of the single nucleotide polymorphism is detected by means of a label associated with the detector primer.
64. (New) The method of Claim 63 wherein the label becomes detectable upon extension of the detector primer or produces a change in signal upon extension of the detector primer.
65. (New) The method of Claim 64 wherein the label is a fluorescent donor/quencher dye pair and a decrease in donor dye fluorescence is detected as an indication of the presence of the single nucleotide polymorphism.
66. (New) The method of Claim 64 wherein a change in fluorescence polarization is detected as an indication of the presence of the single nucleotide polymorphism.

67. (New) The method of Claim 47 wherein the efficiency of detector primer extension is determined quantitatively.

68. (New) The method of Claim 47 further comprising, prior to amplifying, displacing the hybridized detector primer from the target by extension of an upstream primer.

69. (New) A method for detecting a single nucleotide polymorphism in a target sequence comprising, under isothermal conditions at about 37 degrees Celsius:

- a) hybridizing to the target sequence a detector primer comprising a diagnostic nucleotide for the single nucleotide polymorphism which is one to four nucleotides from the 3' terminal nucleotide of the detection primer;
- b) in a primer extension reaction, displacing the detector primer by extension of a second primer hybridized to the target sequence upstream of the detector primer, and;
- c) detecting the presence or absence of the single nucleotide polymorphism based on an efficiency of detector primer extension.

70. (New) The method of Claim 69 wherein the single nucleotide polymorphism is identified using the detector primer.

71. (New) The method of Claim 70 wherein the single nucleotide polymorphism is identified using multiple detector primers, each detector primer comprising a different diagnostic nucleotide.

72. (New) The method of Claim 71 wherein each of the multiple detector primers comprises a different 5' tail sequence.

73. (New) The method of Claim 69 wherein the second primer is an amplification primer.

74. (New) The method of Claim 69 wherein the detector primer comprises a label which becomes detectable upon extension of the detector primer or which produces a change in signal upon extension of the detector primer.

75. (New) The method of Claim 74 wherein the label is a fluorescent donor/quencher dye pair and a decrease in donor dye fluorescence is detected as an indication of the presence or absence of the single nucleotide polymorphism.

76. (New) A method for detecting a single nucleotide polymorphism in a target comprising, under isothermal conditions at about 37 degrees Celsius:

- a) hybridizing a detector primer and a second primer to the target such that extension of the second primer by polymerase displaces the detector primer from the target sequence, wherein the detector primer comprises a diagnostic nucleotide for the single nucleotide polymorphism which is two to four nucleotides from the 3' terminal nucleotide of the detection primer;
- b) extending the detector primer and the second primer with polymerase to produce a displaced detector primer extension product;
- c) determining an efficiency of detector primer extension, and;
- d) detecting the presence or absence of the single nucleotide polymorphism based on the efficiency of detector primer extension.